



**Figure S1. Multi-dimensional analysis identifies peripheral blood monocyte differentiation pathway. Related to Figure 3.** (A) Gating strategy for SPADE analysis of peripheral blood leukocytes. (B-E) Trees for the indicated surface markers colored by median intensity. (F) Flow cytometry analysis of the cells populating each indicated cluster of nodes. Numbers represent percentage of cells from parent gate.

## **Supplemental Experimental Procedures**

## Unsupervised multi-dimensional flow cytometry analysis.

Peripheral blood from Nur77-GFP mice was stained for Lin (B220, Ter119, NK1.1), CD45.2, Ly-6G, Ly-6C, CD11b, CD115, TremL4, CD11c, and MHC-II (I-A/E). Cells were collected on a FACS Aria Fusion (BD) and analyzed with Cytobank software (Cytobank.org, Kotecha et al., 2010). For SPADE analysis, cells were gated as live, singlet, CD45.2<sup>+</sup>. The target number of clusters was set to 200, with density-dependent downsampling to 10%.

## **Supplemental References**

Kotecha, N., Krutzik, P.O., and Irish, J.M. (2010). Web-based analysis and publication of flow cytometry experiments. Curr. Protoc. Cytom. *Chapter 10*, Unit10.